Nitrate Determination by a Modified Conway Microdiffusion Method

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The proposed modified Conway microdiffusion method provides for consecutive determinations of NH_4- and NO_3-N in a given aliquot of soil extract. Analyses of primary nitrate standards showed essentially complete recovery in the range of 1 to 20 ppm NO_3-N (4 to 80 μg N/aliquot). Results for (NH_4+NO_3)-N and NO_3-N in soil extracts are comparable to those obtained, respectively, by macrodistillation with Devarda's alloy and by the phenoldisulfonic acid colorimetric method. The method is rapid and suitable for routine analyses of soil extracts, the equipment is inexpensive, and no interferences are apparent.

Nitrates in soils are derived from mineralization of soil organic nitrogen and from application of nitrogen fertilizers (1). A knowledge of amounts of nitrate present in soils and in drainage waters emanating therefrom may have important implications in guiding judicious management and, hence, minimizing excessive use of nitrogen fertilizers.

Various interferences are encountered in determining nitrate in soil extracts, using colorimetric methods (2) or the nitrate-selective electrode (3). Aqueous extracts of soils and water samples frequently are colored and/or contain suspended clay or organic matter that interfere in colorimetry. Although extraction of soils with aqueous salt solutions (sulfates or chlorides of potassium or calcium are commonly used) flocculates the colloids, extracts often are colored. Chloride interferes with nitrate determinations by the phenoldisulfonic acid colorimetric method and by the nitrate-selective electrode, thus necessitating preliminary removal of chloride or use of compensating chloride solutions in nitrate standards for electrode calibration. These interferences can be avoided by using a modified Conway microdiffusion method.

Despite the foregoing advantages, the microdiffusion method has not tended to replace existing methods for determining nitrates in water and soil extracts. Moreover, the microdiffusion methods most recently reported (2, 4) have not been widely adopted because (*1*) procedures are laborious and time consuming; (*2*) NO_3-N is estimated indirectly as the difference between separate determinations of (NH_4+NO_3)-N and NH_4-N; and (*3*) exacting techniques are required to insure complete reduction of NO_3-N to NH_4-N. The Obrink modification (5) of the Conway microdiffusion unit, in providing a simple and rapid means of sealing the dish from the atmosphere, eliminates an objectionable feature of other units (2, 4). The Obrink modification has been used successfully to measure amounts of NH_4-N in soil extracts (5, 6), and its use for the direct determination of NO_3-N as well as (NH_4+NO_3)-N is described in the present paper.

METHOD

Apparatus

(a) *Microdiffusion dish.*—Obrink modification (5) of Conway microdiffusion dish is obtainable as molded plastic unit consisting of dish and cover (Bel-Art Products, Pequannock, N.J. 07440). Units used in present study (Fig. 1) were ca 83 mm in diameter with sample (No. 2) and acid chamber (No. 1) capacities of ca 6 and 3 ml, respectively. Cover fits peripheral groove or moat (No. 3) which seals unit from atmosphere.

(b) *Buret.*—Micrometer 0.2 ml buret with divisions of 0.2 μl.

Reagents

(a) *Boric acid solution.*—1%. Add 10 g H_3BO_3, 200 ml 95% ethanol, 700 ml distilled water, 10 ml mixed indicator (b), and 0.25 ml NPX Tergitol (nonionic wetting agent, 1+10.5 nonylphenol-ethylene

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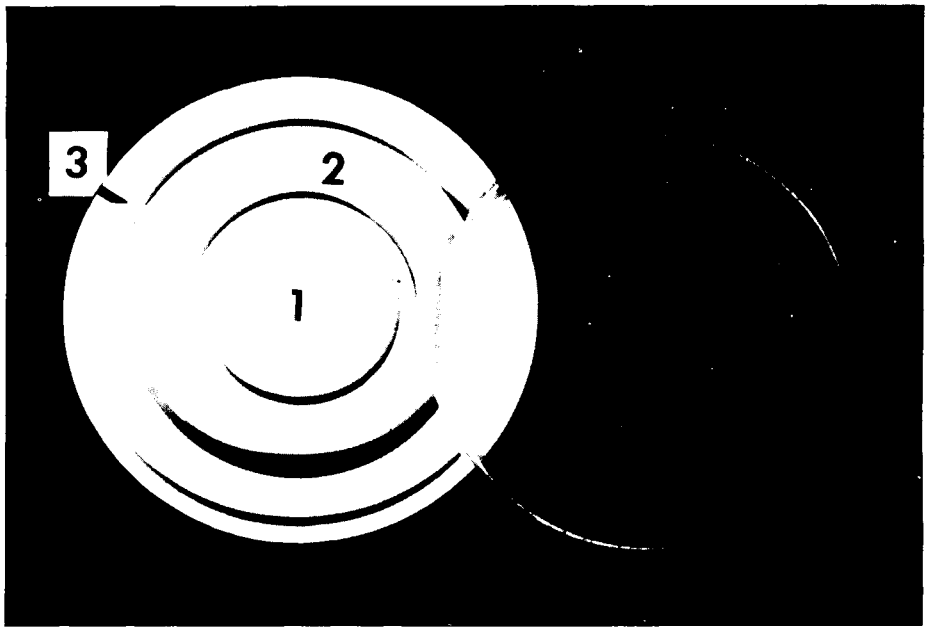

FIG. 1—The Obrink modification of the Conway microdiffusion dish, showing the central boric acid chamber (No. 1), the peripheral sample chamber (No. 2), and the annular moat (No. 3). The clear polypropylene cover, when seated in the moat, seals off the acid and sample chambers.

oxide condensate) to 1 L volumetric flask and let solid dissolve. Cool solution, adjust to faint pink with 0.005*N* NaOH, and dilute to volume with distilled water.

(b) *Mixed indicator.*—Dissolve 0.066 g methyl red and 0.033 g bromocresol green in 100 ml 95% ethanol.

(c) *Devarda's alloy.*—20 mesh or finer (Baker, reagent grade).

(d) *Potassium carbonate solution.*—45% (w/v) in water. Add 4–5 drops of NPX Tergitol/L.

Nitrate Determination

(a) *Microdiffusion.*—Distribute 40–50 mg Devarda's alloy in peripheral sample chamber (No. 2, Fig. 1) of Obrink-Conway dish. Pipet 4 ml solution to be analyzed for nitrate into same chamber. Pipet 1.5 ml boric acid solution into center well (No. 1) of dish. Place 1.5–2 ml 45% K_2CO_3 solution in outer moat (No. 3) and 1 ml in sample chamber. Seat lid in moat. After 16 hr at room temperature, remove lid and titrate boric acid to end point with 0.02*N* H_2SO_4, using micrometer buret (1 ml 0.02 *N* H_2SO_4 titrates 280 μg NH_4-N). Stir solution with buret tip during titration. This procedure measures (NH_4 + NO_3)-N.

To perform consecutive determinations of NH_4- and NO_3-N using single set of sample solutions and microdiffusion units, proceed as follows: First determine NH_4-N by procedure outlined above, omitting Devarda's alloy. After ≥16 hr, titrate diffused NH_4-N. Then distribute Devarda's alloy in sample chamber and replace cover. After ≥16 hr, titrate again to measure NO_3-N. (Because significant amounts of nitrite are rarely found in soil extracts, the present study was limited to NH_4- and NO_3-N. Bremner (2) has described a procedure for estimating NO_2-N in microdiffusion analyses, involving its preliminary destruction with sulfamic acid.)

(b) *Extraction of soil nitrate.*—Several methods commonly are used to extract nitrates from soils (1, 7). The methods used in the present study are briefly described under *Results* in connection with specific experiments.

Results

Nitrate Recovery from Primary Standards.—Nitrate standard solutions prepared from dried potassium nitrate were analyzed by the modified Conway method and results are given in Table 1. Within the range of 1–20 ppm NO_3-N, average recoveries from primary standards were essentially complete. Although coefficients of variation are relatively high for determinations in the 1–2 ppm range, estimates are sufficiently accurate to meet normal requirements in soil and water analyses.

Table 1. Determination of nitrogen in potassium nitrate primary standards by the modified Conway method

NO_3-N concn in std (calcd), ppm	NO_3-N by modified Conway method,[a] ppm	Std dev., ppm	Coeff. of var.,[b] %
1	1.1	0.15	13.5
2	2.1	0.17	8.2
4	4.0	0.10	2.5
8	8.0	0.07	0.8
10	9.9	0.27	2.7
20	19.3	0.45	2.3

[a] Average of 8 determinations.
[b] Coefficient of variation = (standard deviation/mean) × 100; for statistical methods, *see* Snedecor (8).

Microdiffusion vs. Macrodistillation Method.—Nine soils containing a wide range in mineral N contents were extracted with 1*N* KCl (100 g air-dried soil in 250 ml extractant with 4 replicates). After 2–4 intermittent shakings over a half-hour period, the suspension was allowed to settle. Duplicate 4 ml aliquots from the supernatant liquid were analyzed for (NH_4+NO_3)-N by the modified Conway method described earlier. For comparison with a macrodistillation method, additional extract was recovered by filtering. The extract (150 ml), placed in an 800 ml Kjeldahl flask, was analyzed for (NH_4+NO_3)-N by a Devarda's reduction and distillation procedure (5).

Results in Table 2 show reasonably good agreement between amounts of (NH_4+NO_3)-N determined by the microdiffusion and macrodistillation methods. Coefficients of variation were relatively low by both methods. The averages for the micro- and macro-methods, respectively, are based on 8 and 4 determinations.

Microdiffusion vs. Phenoldisulfonic Acid Method.—Soils were sampled from a number of rate-of-N field experiments on sugar beets in Idaho. The samples were ground and mixed, and a portion of each was sent to the Beltsville laboratory for various chemical determinations, including NH_4- and NO_3-N. Soil samples were extracted by heating with 0.01*M* $CaCl_2$ in a steam chest (100°C) for 16 hr as described by Stanford (7). The NH_4-N fraction in the extract was determined by microdiffusion after which Devarda's metal was added and NO_3-N was determined as described under *Method*.

Independent determinations of NO_3-N were made at the Snake River Conservation Research Center (Kimberly, Idaho), using a different method of extraction and the phenoldisulfonic acid colorimetric procedure for determination of NO_3-N. The extractant was a dilute aqueous solution of Ag_2SO_4 (0.17 g/L) and $CuSO_4.5H_2O$ (2.5 g/L). A 50 g soil sample was shaken 10 min with 200 ml extractant, the extract was filtered, and an aliquot was taken for determination of NO_3-N by the phenoldisulfonic acid method, essentially as described by Bremner (2).

Table 2. Comparison of a macrodistillation and a modified Conway method for determining (NH_4+NO_3)-N in soil extracts, using Devarda's alloy as the nitrate reducing agent

Soil series and texture[a]	Macrodistillation[b]		Modified Conway[d]	
	ppm	Coeff. of var.,[c] %	ppm	Coeff. of var., %
Kranzburg sil	24.3	2.5	22.3	4.0
Barnes l	16.7	5.3	15.1	3.2
Aastad cl	34.5	2.9	32.3	3.0
Cecil sl	6.9	3.6	6.6	4.8
Minidoka sil	18.1	5.2	17.5	3.6
Parshall fsl	5.9	6.6	6.2	2.0
Hagerstown sil	16.5	3.9	16.1	4.4
Pullman sicl	41.4	3.6	40.9	2.3
Amarillo fsl	23.0	4.3	22.9	1.2

[a] si = silt; l = loam; c = clay; s = sandy; fs = fine sandy.
[b] Average of 4 separate extractions.
[c] See footnote b, Table 1.
[d] Average of 8 determinations (duplicate of 4 extracts).

Average NO_3-N contents of 122 soils as determined by the colorimetric and microdiffusion methods, respectively, were 12.8 and 12.5 ppm. The relation between NO_3-N (ppm) by the colorimetric method, Y, and NO_3-N (ppm) by the modified Conway method, X, is depicted by the following regression equation (8): $Y = 0.2 + 1.005X$. The coefficient of determination, r^2, was 0.976. Although the overall range in NO_3-N contents of 122 soils extended from approximately 1 to 45 ppm, about 90% of the soils contained 25 ppm or less and 70% contained 15 ppm or less, as shown in Table 3.

Discussion

The determination of NH_4-N and/or NO_3-N by the modified Conway microdiffusion method as outlined in this report has certain advantages over the methods with which it has been compared, from the standpoint of interferences, simplicity, and cost of equipment. The actual time required per NO_3-N determination by an experienced analyst (excluding the diffusion period)

Table 3. Percentage distribution among ranges of NO_3-N contents in 122 soils (ppm NO_3-N based on air-dried soil) as determined by the phenoldisulfonic acid and modified Conway microdiffusion methods

Range in soil NO_3-N contents, ppm	Phenoldisulfonic acid method, %	Modified Conway method, %
0–5	17.2	18.0
5.1–15	54.1	55.7
15.1–25	18.9	18.0
25.1–35	7.4	6.6
35.1–45	2.4	1.7

is about 3 min, or 4–5 min for consecutive determinations of NO_3- and NH_4-N. On the average, the initial sample and reagent transfers consume about 1.5 min, and the titration from 1 to 1.5 min. Since these operations are done on consecutive days, an analyst can average 100 or more determinations per day, in addition to performing the soil extractions and cleaning of Conway dishes and glassware.

The importance of thorough cleaning of the Conway dishes in order to remove the last traces of alkali deserves special emphasis. Routinely, the dishes are flushed under a tap water faucet while the Devarda alloy residue is removed by brushing. Dishes then are soaked in a vessel of tap water containing detergent, followed by soaking in a dilute acid bath. Finally, the dishes are rinsed thoroughly in tap water, given a final rinse with distilled water, and drained. The lids are rinsed thoroughly in tap water and finally with distilled water.

A unique feature of the modified microdiffusion method as proposed herein is that it provides for the direct determinations of NH_4- and NO_3-N. In the method described by Bremner (2), involving solutions containing both mineral forms of N, separate determinations are made for NH_4-N and for (NH_4+NO_3)-N, and NO_3-N is obtained by difference. The proposed direct method involves an initial determination of diffusible NH_4-N, followed by addition of Devarda's alloy and determination of NO_3-N. Bremner and Shaw (4) concluded that Devarda's alloy used with strong alkalies (potassium carbonate and potassium hydroxide) was unsatisfactory for determining nitrates in soil extracts. They proposed the use of titanous sulfate with MgO-suspension for nitrate reduction in microdiffusion analyses, a method which entails mixing to minimize gel formation and facilitate diffusion of NH_3 (2). Results of the present study, however, show satisfactory recovery of NO_3-N from standards and good agreement with values obtained by the macrodistillation and phenoldisulfonic acid methods.

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